

molecular informatics

Supporting Information

ACE2-Variants Indicate Potential SARS-CoV-2-Susceptibility in Animals: A Molecular Dynamics Study

Szymon Pach⁺, Trung Ngoc Nguyen⁺, Jakob Trimpert, Dusan Kunec, Nikolaus Osterrieder,* and Gerhard Wolber* © 2021 The Authors. Molecular Informatics published by Wiley-VCH GmbH. This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

Supporting Information

ACE2-Variants Indicate Potential SARS-CoV-2-Susceptibility in Animals: An Extensive Molecular Dynamics Study

Szymon Pach^{†[a]}, Trung Ngoc Nguyen^{†[a]}, Jakob Trimpert^[b], Dusan Kunec^[b], Nikolaus Osterrieder^{*[b]}, Gerhard Wolber^{*[a]}

ORCID IDs: Szymon Pach: 0000-0001-6109-7123, Trung Ngoc Nguyen: 0000-0002-7415-4390, Jakob Trimpert: 0000-0003-1616-0810, Dusan Kunec: 0000-0001-6697-7621, Nikolaus Osterrieder: 0000-0002-5313-2176, Gerhard Wolber: 0000-0002-5344-0048

[a] *Pharmaceutical and Medicinal Chemistry, Institute of Pharmacy, Freie Universität Berlin
Königin-Luise-Str. 2 - 4, 14195 Berlin, Germany*

[b] *Institut für Virologie, Freie Universität Berlin
Robert-Von-Ostertag-Str. 7 - 13, 14163 Berlin, Germany
e-mail: Gerhard Wolber (gerhard.wolber@fu-berlin.de), Nikolaus Osterrieder (no.34@fu-berlin.de)

[†] *These authors contributed equally.*

Table S1: Principle component (PC) analysis of eight selected molecular descriptors.

Feature	Eigenvalue	Variance [%]
PC 1	1.93	24.1
PC 2	1.67	20.8
PC 3	1.27	15.9
PC 4	1.07	13.3
PC 5	0.74	9.2
PC 6	0.62	7.8
PC 7	0.40	5.0
PC 8	0.32	3.9

Feature	Depth	Bp C			Bp B		Bp A	
		Deformation	χ^1 Angle ACE2- F/Y82/83	Lipophilic contacts S-F486 – ACE2- F/Y82/83	Distance S-Q493 – ACE2- residue 30/31	Distance ACE2- residues 30/31 – 34/35	Distance S-K417 – ACE2- residue 29/30	Hydrogen bond count
PC 1	12,9%	2,4%	6,5%	5,5%	26,9%	31,0%	6,5%	8,2%
PC 2	18,8%	0,1%	24,5%	23,2%	2,7%	0,5%	13,6%	16,6%
PC 3	0,1%	30,6%	24,0%	1,3%	16,5%	7,8%	8,7%	10,9%
PC 4	21,2%	44,5%	0,0%	0,2%	2,3%	7,2%	20,1%	4,4%
PC 5	13,1%	0,5%	1,2%	64,1%	4,1%	4,3%	6,7%	6,0%
PC 6	0,4%	1,8%	0,0%	0,1%	0,0%	0,8%	43,1%	53,8%
PC 7	6,6%	12,9%	18,0%	0,5%	26,8%	34,2%	0,8%	0,2%
PC 8	26,9%	7,1%	25,7%	5,2%	20,6%	14,1%	0,5%	0,0%

Abbreviations: ACE2- Angiotensin-converting enzyme 2, Bp- binding pocket, PC- principle component, S- spike. Red numbers represent the highest value in a row, the orange numbers the second-highest value.

Human	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEEQAKTFLDKFNHEADLFYQSSLASWNYNTNITEENVQNMNAGDKWSAFLEQSTLAQHYPQETQNLTVKLQALQQSGSVLSADKSKRLNT
Dog	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	QSTEDLVKTFLEKFNFEAEELSYQSSLASWNYNTNITEENVQNMNAGDKWSAFYEEQSKLAKTYPLEETQDSTVVRQLRALQSGSVLSADKNQRLNT
Cat	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEELAKTFLKFNHEAEELSYQSSLASWNYNTNITEENVQNMNAGDKWSAFYEEQSKLAKTYPLAEIHTNTVVRQLRALQSGSVLSADKSKRLNT
Ferret	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEDLAKTFLKFNFEAEELSYQSSLASWNYNTNITEENVQNMNAGDKWSAFYEEQSHAKTYPLEEQDPIIKRQLRALQSGSVLSADKSKRLNT
Syrian hamster	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEEQAKTFLDKFNQEAEDLSYQSSLASWNYNTNITEENAKMNEAAAKWSAFYEEQSKLAKNYSLQEVQNLTIKRQLRALQSGSVLSADKSKRLNT
Mouse	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEENAKTFLNNFQEAEDLSYQSSLASWNYNTNITEENAKMSEAAAKWSAFYEEQSKTQSFSLQEQITPIIKRQLRALQSGSVLSADKSKRLNT
Rat	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	SLIEEKAESFLNFKQEAEDLSYQSSLASWNYNTNITEENAKMNEAAAKWSAFYEEQSKIAQNFSLQEQITQATIKRQLRALQSGSVLSADKSKRLNT
Red squirrel	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEESAKTFLDKFNQEAEDLSYQSSLASWNYNTNITEENAKMNEAAAKWSAFYEEQSKLAKTYPLQIQNLTVKRQLRALQSGSVLSADKSKRLNT
Chinese hamster	20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEEQAKTFLDKFNQEAEDLSYQSSLASWNYNTNITEENAKMNEAAAKWSAFYEEQSKLAKNYSLQEVQNLTIKRQLRALQSGSVLSADKSKRLNT
Campbell's dwarf hamster	10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115	STIEEQAKTFLDKFNQEAEDLSYQSSLASWNYNTNITEENAKMNEAAAKWSAFYEEQSKLAKNYPQDQVNLTIKRQLRALQSGSVLSADKSKRLNT
Human	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWESHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVNGDVG
Dog	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNSMTIYSTVGKACNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEENGYN
Cat	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNAMSTIYSTGKACNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHAKSKQVNYEDYGDYWRGDEYVEETDGYN
Ferret	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNAMSTIYSTGKACNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEEADGYN
Syrian hamster	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Mouse	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Rat	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Red squirrel	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Chinese hamster	120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Campbell's dwarf hamster	115 120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215	ILNTHSTIYSTGKVCNPNPQELLLEPLGLDINHANSIDYNERLHAWEGHRSVEGKQLRPLVEEYVVLKNEHARAN--HYEDYGDYWRGDEYVEADGYN
Human	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YSRQLIEDVEHTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Dog	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YSRQLIEDVEHTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Cat	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YSRQLIEDVEHTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Ferret	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YSRQLIEDVEHTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Syrian hamster	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Mouse	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Rat	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Red squirrel	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Chinese hamster	220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Campbell's dwarf hamster	210 215 220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315	YNGQLIEDVERTFEEIKPLYEHLHAYVRKLMNAPYSISPTGCLPAHLGDMHGRFNTNLYLTPVFGQKPNIDVDAMVQNDARRIFKEAEKFFV
Human	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Dog	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Cat	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Ferret	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Syrian hamster	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Mouse	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Rat	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Red squirrel	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Chinese hamster	320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Campbell's dwarf hamster	310 315 320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415	SVGLPNNITGQFNHNSLTDPGNVQKAVCHPTAWDLGKDFRIKMTKVTMDNFLTAHHEMGHIQYDMAYAPQFLLRNGANEGHEAVGEIMSLSAATPK
Human	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Dog	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Cat	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Ferret	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Syrian hamster	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Mouse	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Rat	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Red squirrel	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Chinese hamster	420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Campbell's dwarf hamster	410 415 420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515	HLKSTIGLSPDFQEDNETEINFLKQALITVGLPFTYHLEKRNHNVFGEIPKQNMKNWENKREIVGVVEPLPHDETICYPASLFHVSNDYSFIRYV
Human	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Dog	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Cat	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Ferret	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Syrian hamster	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Mouse	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Rat	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Red squirrel	520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Chinese hamster	510 515 520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD
Campbell's dwarf hamster	510 515 520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615	TRTIYQFQFQALQAAKHGGLHKKDISNTEAGQKLLNMLRGKSEPTLALENVVGAKNHNRPLLNYPFLFTWLKQNNKNSFVGNHTDWSPYAD

Figure S1: Sequence alignment of human and animal angiotensin-converting enzyme 2 performed with MOE 2019.0102 (Molecular Operating Environment, Chemical Computing Group ULC, Montreal, Canada). Colour code: yellow box- cysteine residues involved in a disulfide bridge.

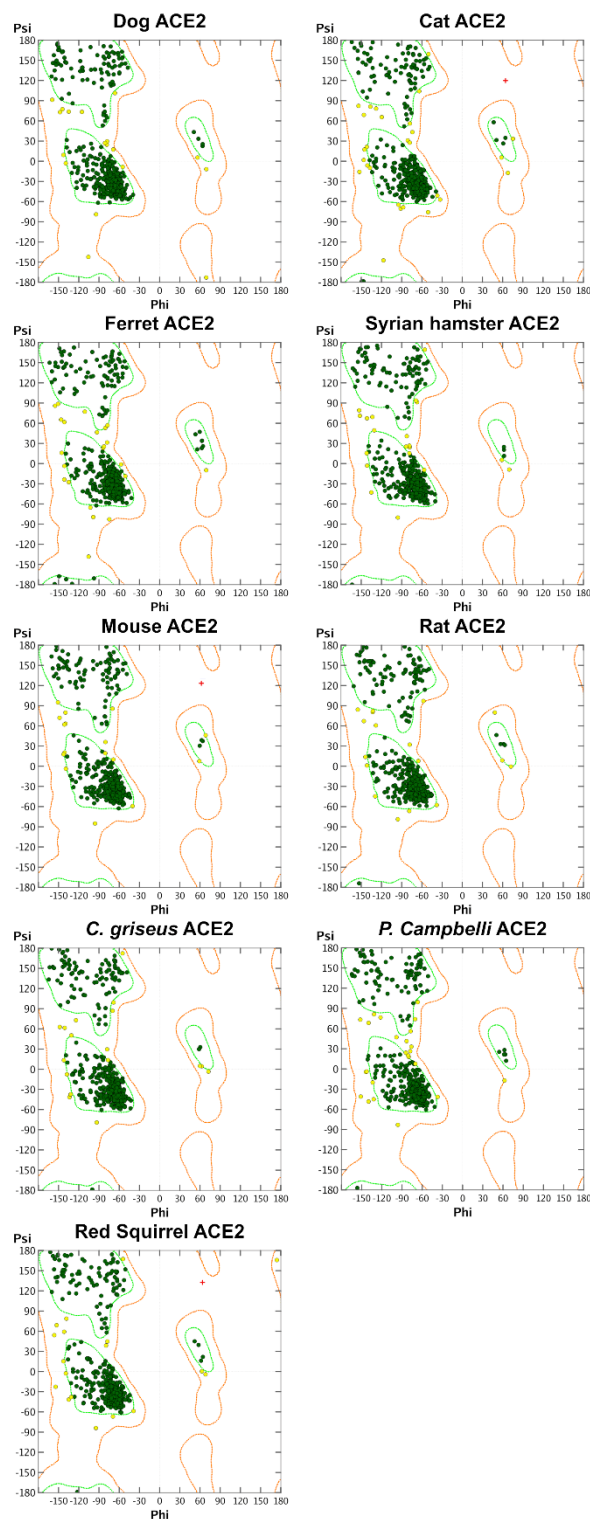


Figure S2: Ramachandran plots for all ACE2 homology models. Symbol code: green point- residue with favourable geometry, yellow point- residue with allowed geometry, red cross- outlier. The figure was generated using MOE 2019.0102 (Molecular Operating Environment, Chemical Computing Group ULC, Montreal, Canada).

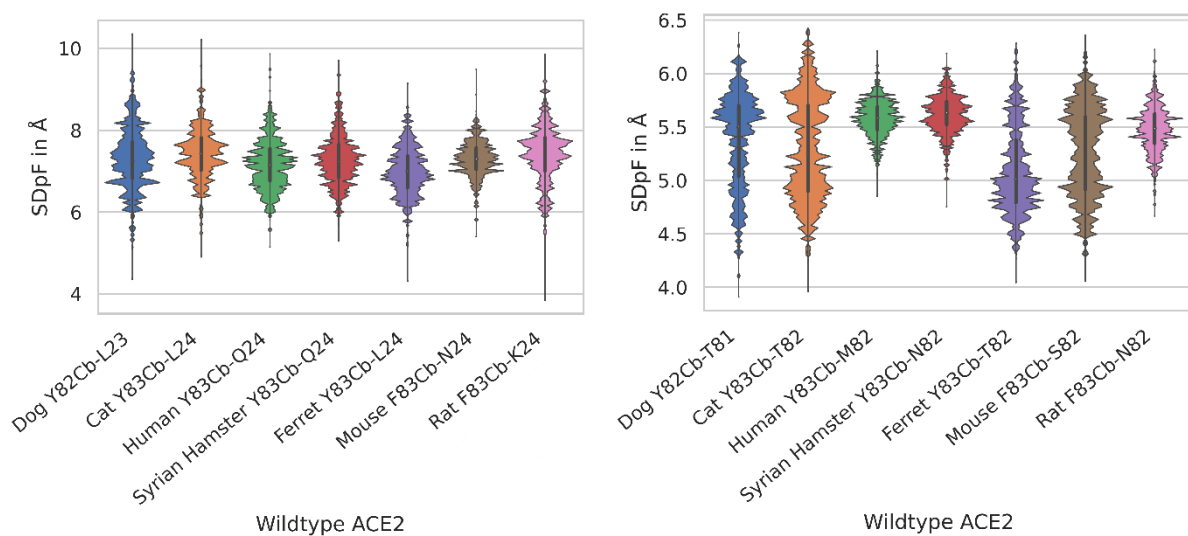


Figure S3: Distance distribution between Bp C key residues for analyzed wildtype ACE2 species. Shortest Distance per Frame (SDpF) between the Cb atom of residue 83 (or 82 in dog) and side chains flanking the binding pocket C (residues 24 and 82 or 23 and 81 in dog) representing the 'depth' of Bp C.

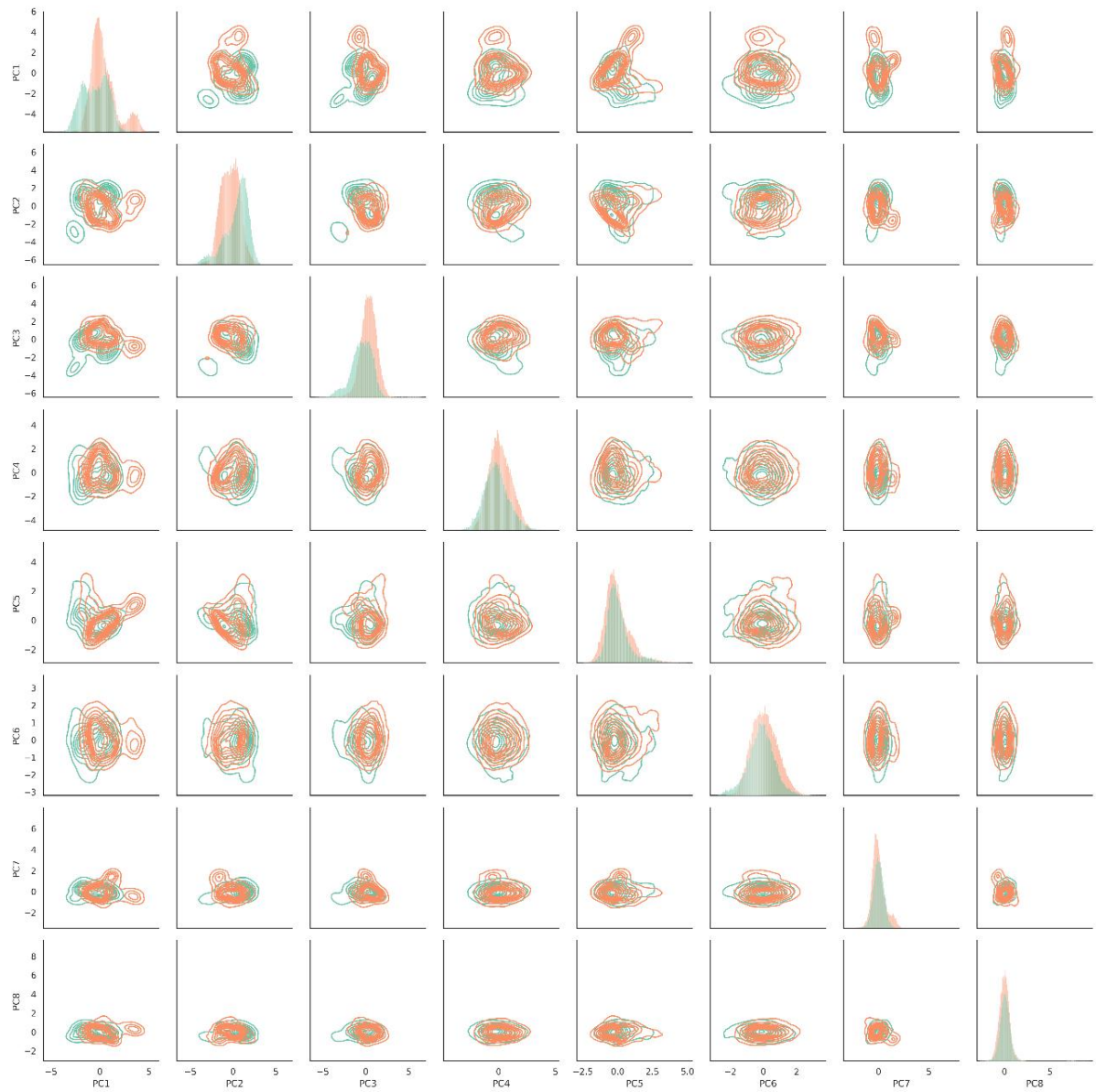
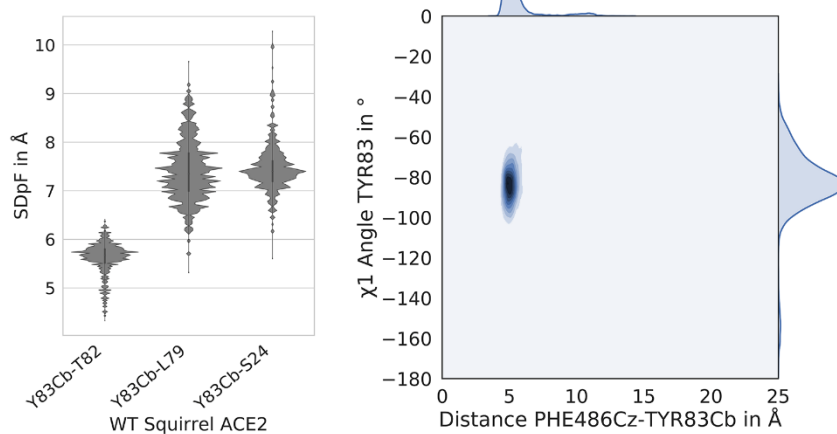
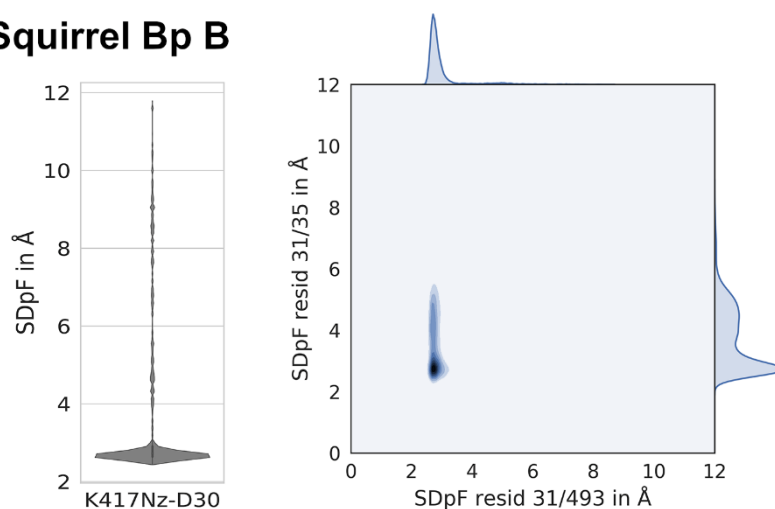


Figure S4: Histograms of principle components (PC) and kernel density plots of PC pairs visualizing the variance dependent ability of our feature set to discriminate between susceptible (orange lines) and non-susceptible species (green lines).

Squirrel Bp C



Squirrel Bp B



Squirrel Bp A

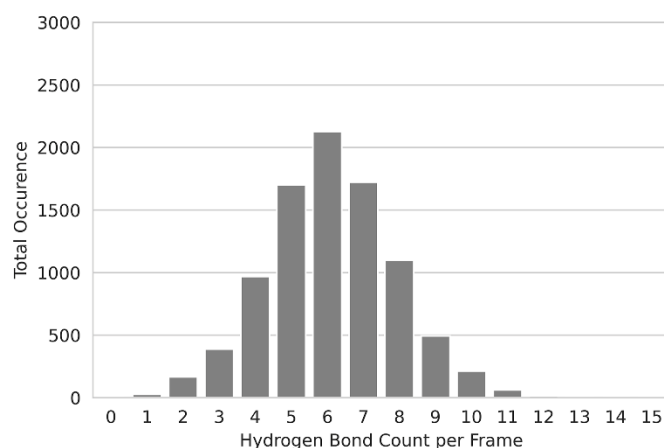
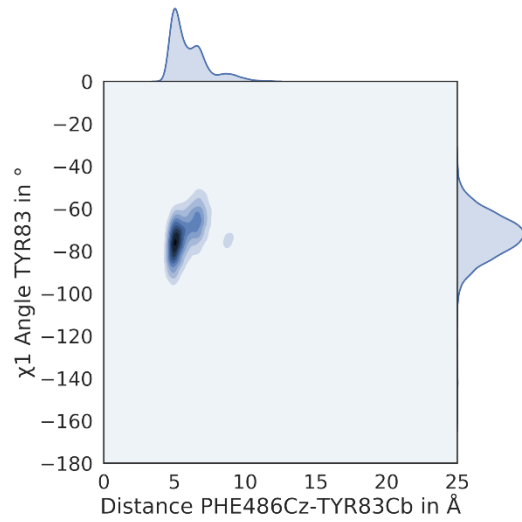
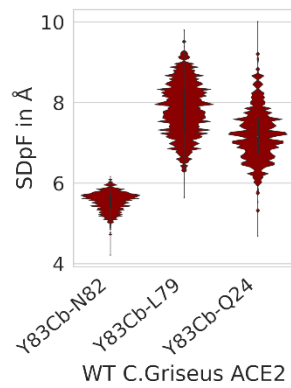
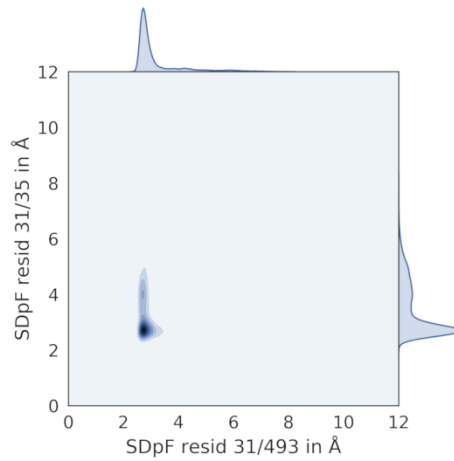
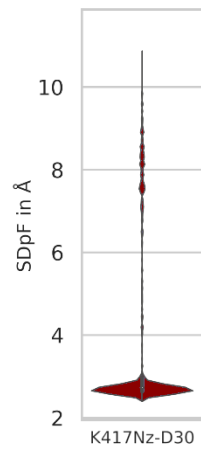


Figure S5. Descriptors calculated for red squirrel ACE2-RBD simulations. **Bp C:** Shortest Distance per Frame (SDpF) between Cb atom of Y83 and side chains of residues flanking the binding pocket; **Bp B:** Distance distribution for interactions between Nz atom of RBD K417 and the side chain of ACE2 residue 30, and kernel density plots summarizing the interactions between ACE2-residue 31 and RBD residue Q493; **Bp A:** histograms representing the total occurrence of hydrogen bonds in binding pocket A between ACE2 residues 37, 38, 41, 42, 353, 355 and RBD residues 449, 496, 498, 500, 501, 505.

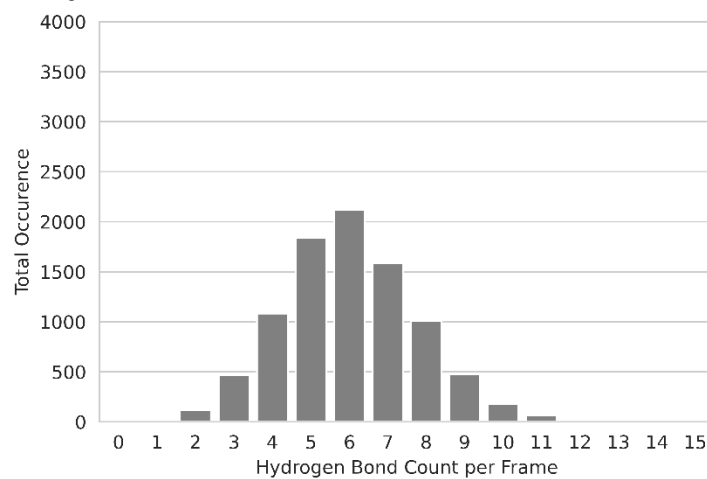
***C. griseus* Bp C**



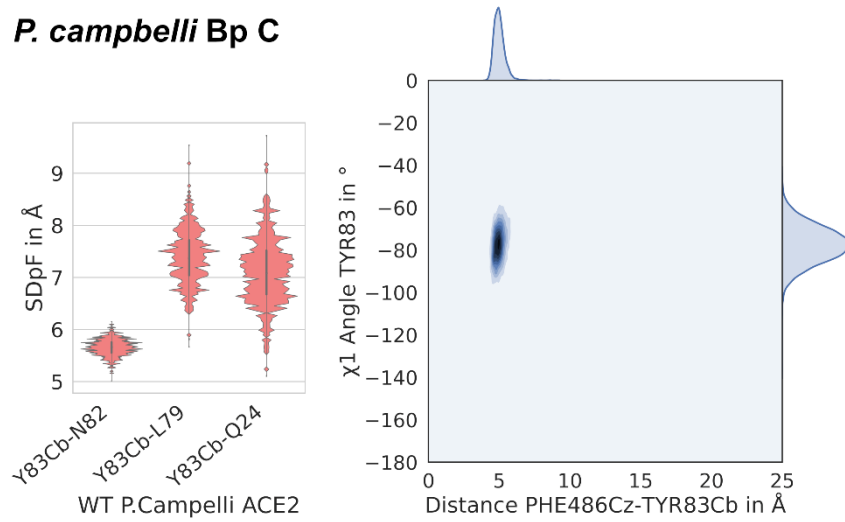
***C. griseus* Bp B**



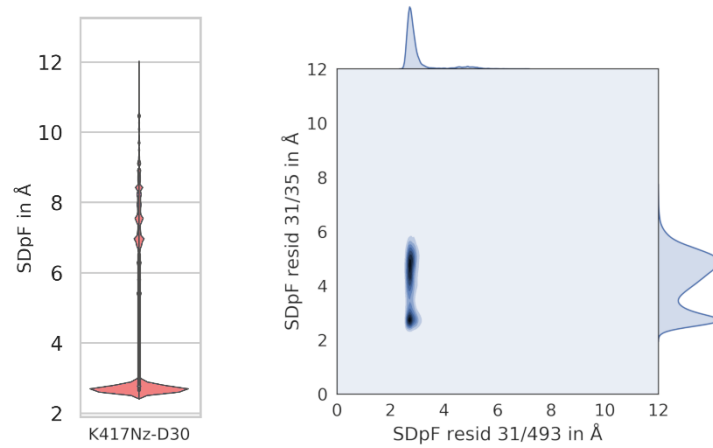
***C. griseus* Bp A**



P. campbelli Bp C



P. campbelli Bp B



P. campbelli Bp A

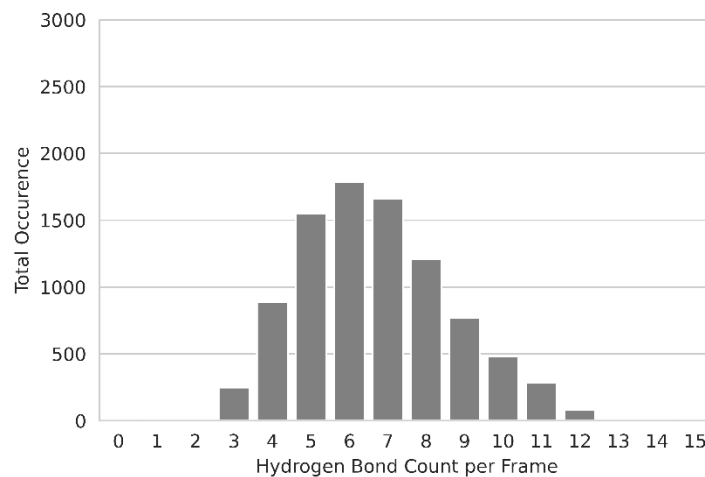


Figure S6. Descriptors calculated for small hamsters (*C. griseus*, *P. campbelli*) ACE2-RBD simulations. **Bp C:** Shortest Distance per Frame (SDpF) between Cb atom of Y83 and side chains of residues flanking the binding pocket; **Bp B:** Distance distribution for interactions between Nz atom of RBD K417 and the side chain of ACE2 residue 30, and kernel density plots summarizing the interactions between ACE2-residue 31 and RBD residue Q493; **Bp A:** histograms representing the total occurrence of hydrogen bonds in binding pocket A between ACE2 residues 37, 38, 41, 42, 353, 355 and RBD residues 449, 496, 498, 500, 501, 505.

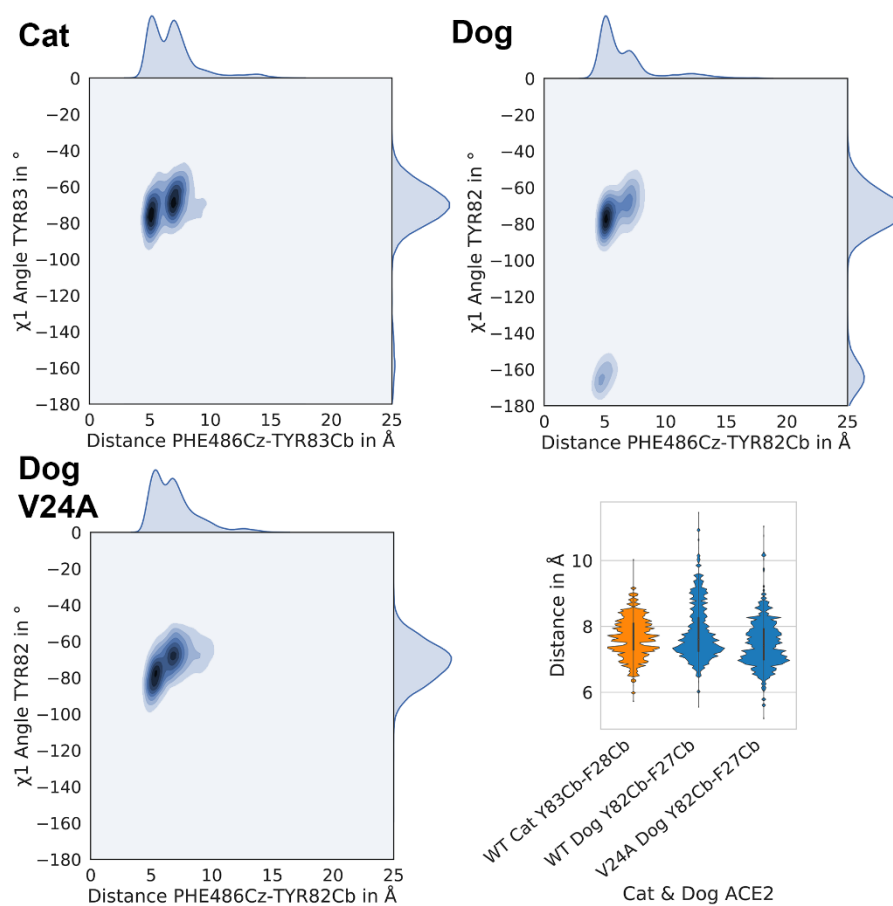


Figure S7. Occupation of binding pocket C indicated by kernel density plots (x-axis: distance F486 Cz - Y82 Cb; y-axis: χ_1 angle of central residue 82/83) and Bp C deformation of in dog ACE2 wild type, V24A mutant, and homologous residue 83 in cat (Y83/82 Cb – F28/27 Cb distance).

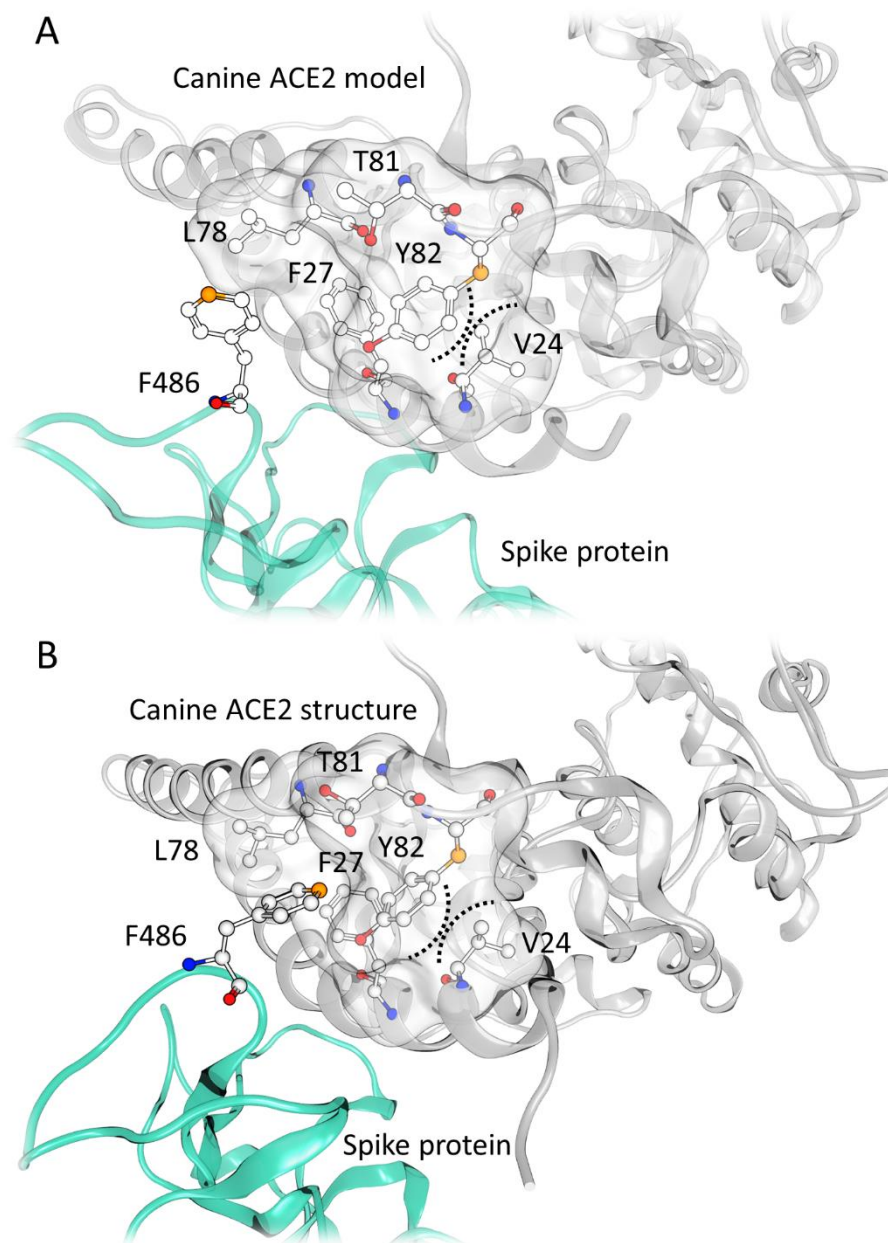


Figure S8. Comparison of (A) the “fixed state” in our model of canine angiotensin-converting enzyme 2 (ACE2) in complex with viral spike protein extracted from a molecular dynamics simulation and (B) the cryoscopic electron microscopy structure of the dog ACE2 in complex with the viral spike protein.

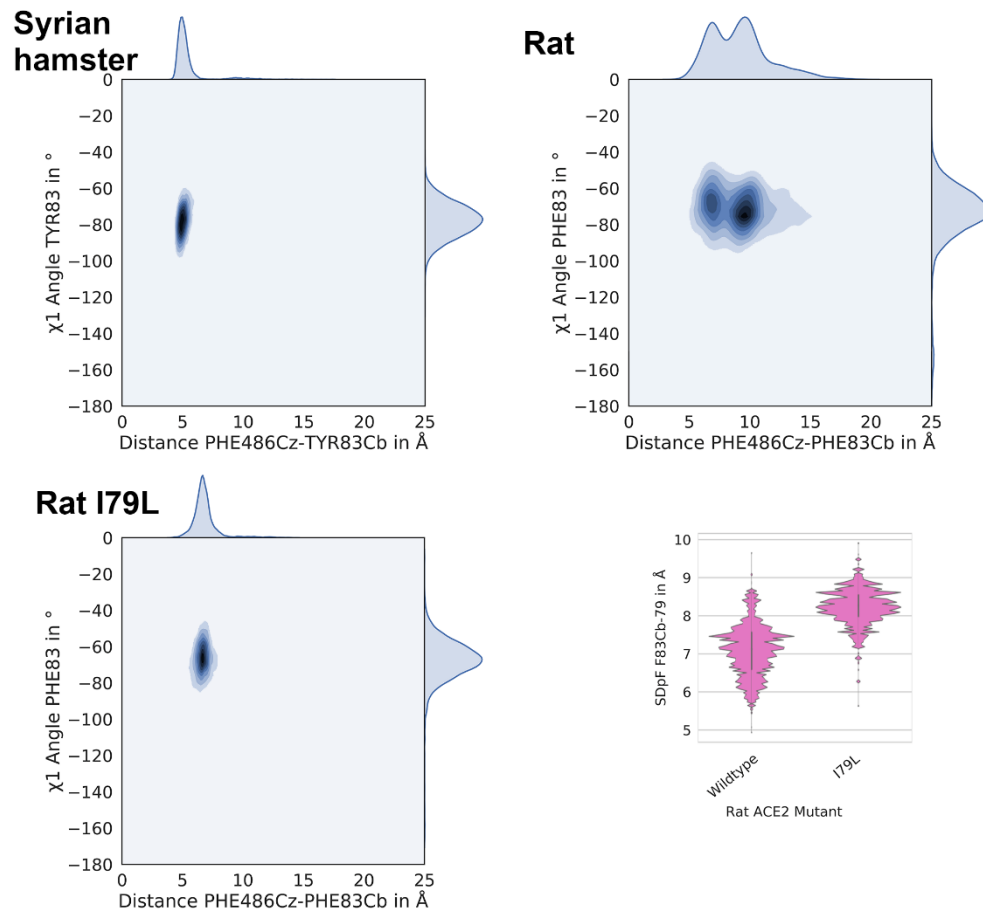
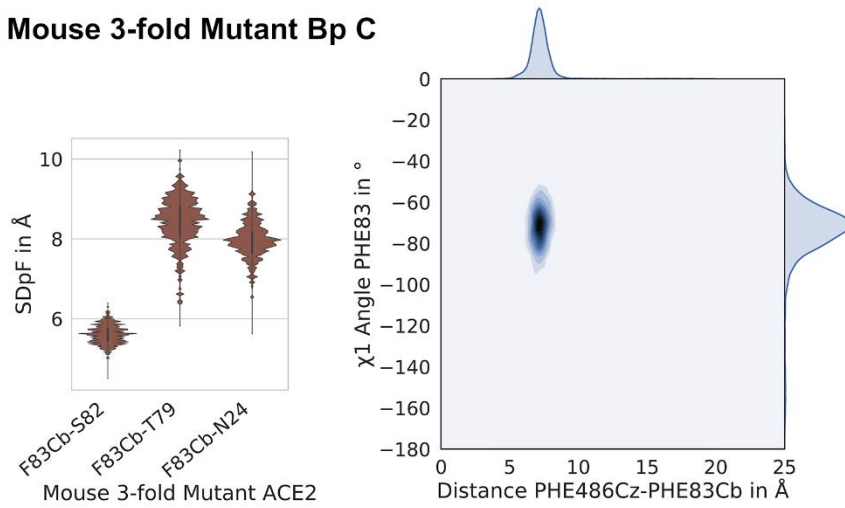
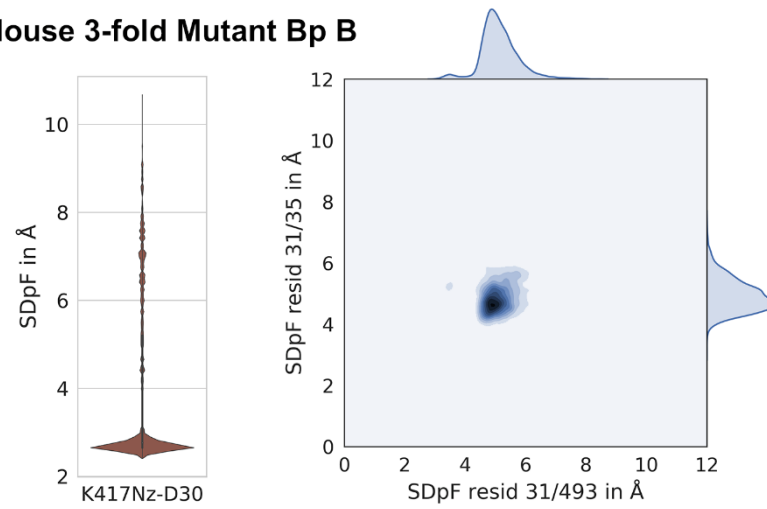


Figure S9. Kernel density plots summarizing the occupation of binding pocket C (x-axis, surrogate parameter: distance F486 Cz - Y83 Cb) and rotamers of central residue 82 (or 83 in cats). The lower right panel shows the opening of the Bp C based on the SDpF-83-79 (Shortest Distance per Frame) parameter.

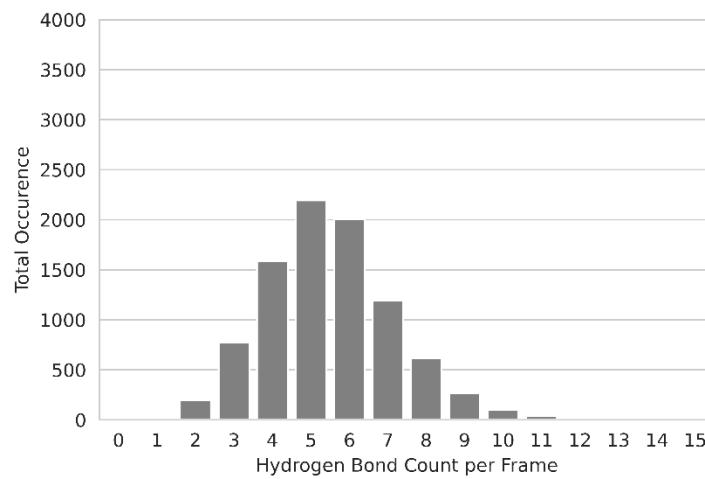
Mouse 3-fold Mutant Bp C



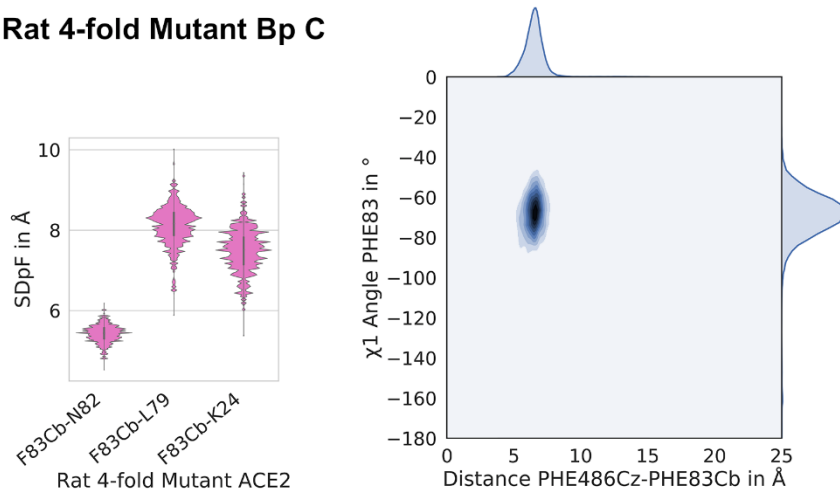
Mouse 3-fold Mutant Bp B



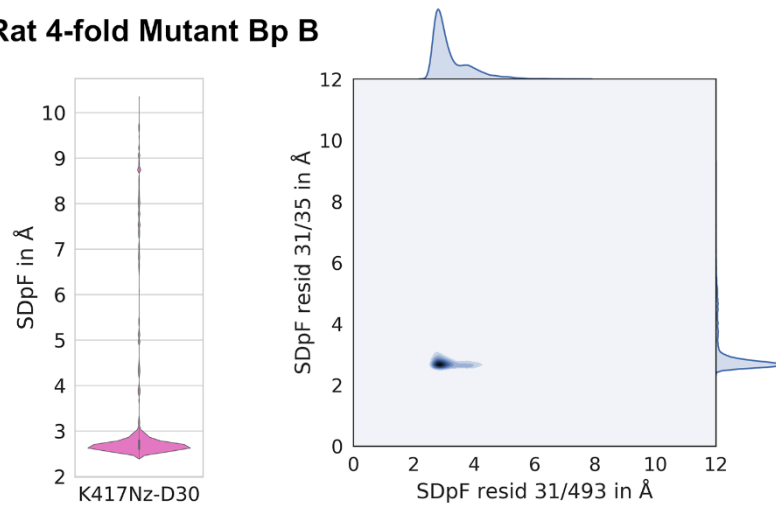
Mouse 3-fold Mutant Bp A



Rat 4-fold Mutant Bp C



Rat 4-fold Mutant Bp B



Rat 4-fold Mutant Bp A

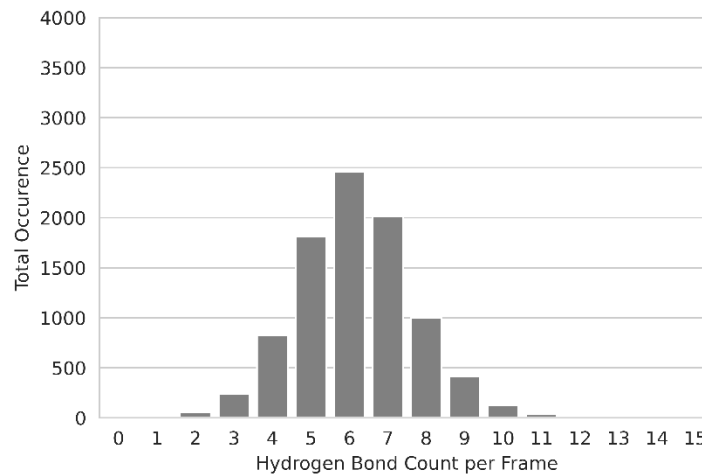


Figure S10. Descriptors calculated for mouse triple mutant (upper panels) and rat quadruple mutant (lower panels) ACE2-RBD simulations. **Bp C:** Shortest Distance per Frame (SDpF) between Cb atom of Y83 and side chains of residues flanking the binding pocket; **Bp B:** Distance distribution for interactions between Nz atom of RBD K417 and the side chain of ACE2 residue 30, and kernel density plots summarizing the interactions between ACE2-residue 31 and RBD residue Q493; **Bp A:** histograms representing the total occurrence of hydrogen bonds in binding pocket A between ACE2 residues 37, 38, 41, 42, 353, 355 and RBD residues 449, 496, 498, 500, 501, 505.